Response of the germinable soil-stored seed bank of a remnant reserve in the southern Western Australia agricultural zone to smoke and fire treatment

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Abstract

The seedling emergence method was used to germinate soil samples obtained from East Yornaning Nature Reserve in the agricultural wheatbelt of Western Australia. Four treatments (control, aerosol smoke, aqueous smoke and fire) were used to assess germination response under greenhouse conditions. Emergence was monitored on a weekly basis for six months and seedlings classed according to life form (grass, sedge, herb or woody perennial) and into native and non-native categories. The species composition of the soil seed bank was dominated mainly by native herbaceous plants with few woody perennials, unlike the standing vegetation. There was a significant promotive effect of aerosol smoke on seedling germination (102 seedlings / m^2) relative to the control (43 seedlings / m^2). Non-native seedlings such as *Hypochaeris* sp., *Arctotheca calendula, Sonchus* sp. and *Ursinia anthemoides* represented between 5% (control treatment) and 12% (fire treatment) of total seedling emergence. Understanding the abundance and dynamics of species in the soil-stored seed bank can provide assistance for management of small remnant vegetation communities in the event of disturbance such as wildfire.

Keywords: soil-stored seed bank, non-native and invasive plants, Western Australia, fire.

Introduction

The soil-stored seed bank is the genetic resource for restoration or maintenance of plant communities should the standing vegetation be adversely disturbed (Roberts 1981; Fenner 1995). Many species killed by fire rely on a soil-stored seed bank to recruit individuals following fire (Bell 1999). Knowledge of the abundance and dynamics of species assemblages in the soil seed bank are important for predicting the outcome of different management strategies (Noble & Slatyer 1980; Keddy *et al.* 1989) in addition to providing information on the susceptibility of local plant communities to non-native plant invasion.

In fire-prone habitats many species with soil-stored seed banks have evolved barriers to germination that are overcome by fire-related cues, such as smoke and/or heat (Bell 1999). In Western Australia, with its Mediterranean climate of hot dry summers and cool wet winters, fire is a frequent phenomenon and is often used as a management tool. For these purposes it is critical to have a good understanding of the effect of fire, and smoke, on natural ecosystems.

Disturbance events, such as habitat fragmentation and fire, can facilitate the invasion of native ecosystems by alien plants and cause negative effects for the regeneration of native biodiversity (Groves & Willis 1999, Abensperg-Traun *et al.* 1998, Drake 1998). Non-native species can be defined as those that have been introduced from elsewhere. Many are widespread generalists that often display non-specialised habitat requirements, long distance seed dispersal and/or long lived soil-stored seed banks. Many also respond to nutrient increases as occurs after fire. Many have the potential to become weeds through rapid colonization of disturbed areas (Hussey *et al.* 1997). They may pose a major problem to management of native plant communities and may threaten the continued survival of many native species (Groves & Willis 1999). The most successful of these weedy species are those with attributes such as long distance dispersal mechanisms or long-lived seed banks (Thompson 1992). Non-native plant species may compete above and below ground for nutrients, water, light and space (Lenz & Facelli 2005) and interrupt ecosystem processes such as nutrient and water cycling (Holmes 2002). In addition, the flammability of a habitat may increase after invasion by non-native plant species, especially grasses, due to higher fuel loading (Abensperg-Traun *et al.* 1998, Thomson & Leishman 2005), which can in turn lead to changes in the intensity of future fires or increased fire frequency (Milberg & Lamont 1995, Holmes 2002).

The aim of this study was to investigate the composition of the soil-stored seed bank of a long unburnt nature reserve in the southern agricultural zone. To determine the germination response of the soil seed bank the seedling emergence method was used in conjunction with a number of treatment conditions designed to emulate disturbance events. With more than 65% of this zone cleared for agriculture small patches of remnant vegetation are of particular concern for

Figure 1. East Yornaning Nature Reserve

conservation. The responses of the seedbank to the varied treatments will have implications for management and conservation of small reserves.

Materials and Methods

Study Site

East Yornaning Nature Reserve is located approximately 30 km northeast of Narrogin, Western Australia (Latitude 32° 47' 06", Longitude 117° 17' 11") (Fig.1). This 248 hectare remnant in the southern agricultural wheatbelt of the State is long unburnt (~45 years) and contains the only substantial population of the endangered *Acacia insolita* E. Pritz. subsp. *recurva* Maslin. The study site was located on a laterite breakaway where this rare *Acacia* is situated in association with open woodland and shrubland of *Allocasuarina huegeliana, Xanthorrhoea preissii, Eucalyptus wandoo, Dryandra nivea* subsp. *nivea* and *D. nobilis.* Many of the woody perennial species present at the site are obligate seeders, including members of the Mimosaceae and Papilionaceae. *E. wandoo* and *X. preissii* have the ability to resprout after fire. Several species, like *A. huegeliana, E. wandoo* and the *Dryandra* species, store seed in their canopy, in some cases for many years. The study site was situated centrally within the reserve, approximately 200 m from the nearest boundary with agricultural land.

Experimental Design

Soil samples were collected in autumn 1998 after most species had completed fruiting and had released seeds. Eight plots, each measuring 5 m^2, were randomly chosen along the length of the laterite breakaway in the reserve and confined within the bounds of the rare *A. insolita* subsp. *recurva* population. Ten randomly selected soil samples (25 cm x 25 cm x 5 cm) were collected from each plot and samples from each plot were bulked. These eight bulked samples were air-dried and passed through a series of different mesh-sized sieves to remove twigs and break up clay particles to ensure friability of the soil. During this process all *A. insolita* subsp. *recurva* seeds were removed from the sample. Further investigations into the *Acacia* are to be described separately (Cochrane *et al.* in prep) and these seeds have not been taken into consideration in this present investigation.

The seedling emergence method was used to determine presence and abundance of different life forms present in the soil-stored seed bank. Soil from the eight plots was divided evenly into 35 cm x 29 cm plastic nursery trays. Trays from each plot were randomly assigned to one of three treatments or to a control (no treatment), such that each treatment had eight trays, one from each of the eight plots. All trays were watered with the soil wetting agent Wettasoil[HG] prior to treatment to eliminate differences in wetting capacity. The soil was then thoroughly drenched to ensure even wetting. Smoke treatment 1 was administered by aerosol smoking of trays for 60 minutes in a tent. All smoke treatment 1 trays were covered with plastic for six days after treatment to prevent leaching of smoke chemicals from trays before seed had the ability to imbibe the chemicals from the smoke. Smoke treatment 2 was administered by applying aqueous application of smoke onto trays (smoke generated by burning native vegetation bubbled through water for 60 min and poured onto trays at 500 ml / tray). A low intensity fire treatment was administered by placing leaf litter on trays to a depth of two centimetres and ignited. The control received no treatment. After the treatments were applied all trays were randomly arranged in a shade house. Two extra trays of sterilised soil were also placed in the shade house to determine windblown seed rain.

Seedling emergence and growth were monitored on a weekly basis. All germinating seedlings were marked with a toothpick to prevent recounting. When seedlings were sufficiently large enough for identification into life forms (grass, sedge, herb or woody perennial) and native and non-native status they were carefully removed from trays. Where possible, seedlings were identified to genus level. Monitoring of the soil seed bank in the shade house continued for six months until it was assumed that all seed had germinated or died, or remained dormant under the conditions provided during the study.

Statistical analysis

Seedling count data have been presented as the density of seedlings per m^2. A one-way analysis of variance (ANOVA) (STATVIEW for Windows Version 5.0.1) determined differences between treatments, life forms and plots for total seedling emergence at three and six months. A two-way ANOVA determined the interaction between treatment and life form. Pairwise comparisons were made with Fishers PLSD to determine significant differences between treatment and life forms.

Results

Species that germinated from the soil seed bank were allocated to groups based on their life form (Table 1). Seeds of native grasses, sedges, herbs and woody perennials, as evidenced by their seedlings, were all represented in the soil seed bank, although the numbers of herbaceous plants (for example, *Trachymene* sp., *Wahlenbergia* sp., *Levenhookia* sp., *Phyllanguim* sp. and members of the family Asteraceae) that emerged was significantly greater than for any other life form ($P<.0001$) (Table 1). The few woody perennials that did germinate included *Allocasuarina* sp., *Eucalyptus* sp., *Dryandra* sp., *Adenanthos* sp. and a member of the Papilionaceae, possibly *Gastrolobium* sp.

Significantly more seedlings emerged within the first three months ($P < 0.0001$) (67%) accounting for 868 seedlings or 173 seedlings / m^2. A further 413 seedlings emerged in the second 3-month period, bringing total seedling emergence over all plots and treatments to 1281 (256 seedlings / m^2) (Table 1). The proportion of seedlings of each life form that germinated at three and six months relative to total seedling emergence was similar for each time period (Table 1).

Treatment enhancement of life forms

Smoke applied in aerosol form enhanced germination for all life forms except grasses ($P < 0.05$) (Fig. 2). Native grasses required no stimulatory treatment for germination, with 30% of all grasses (52 out of 150) germinating in the control. Herbaceous perennials showed the most significant increase in germination with aerosol smoke treatment (Fig. 2). A total of 219 seedlings (43 seedlings / m^2) emerged from untreated soil during the study. When aerosol smoke was applied to soil, 513 seedlings emerged (102 seedlings / m^2). Both aqueous smoke application and the burning of litter on the soil surface also increased seedling emergence over the control (289 and 260 seedlings respectively), but not significantly (P = 0.6141 and 0.7677). The rate of emergence between treatments was similar and ranged from 57% in 3 months in the aqueous smoke treatments to 73% in the aerosol smoke treatment. 68% of seedlings emerged within 3 months in the control.

Non-native versus native seedling emergence

Common non-native species such as *Hypochaeris* sp., *Arctotheca calendula* and *Ursinia anthemoides* were also present in the soil seed bank representing 8% of all seedlings that emerged within six months (20 seedlings / m^2) (Table 1). There was a significant difference between non-native and native seedling emergence between treatments (P = 0.023), with the greatest number of non-native seedlings (35) occurring in the aerosol smoked treatment (Fig. 3). Emergence of non-native plant species in control, aerosol smoke, aqueous smoke and fire treatments as a proportion of the total number of seedlings emerging in each treatment was 5%, 7%, 9% and 12% respectively.

Table 1

Seedling emergence by life form and their relative contribution to recruitment after disturbance determined from soil seed bank analysis after three and six months using the seedling emergence technique under shade house conditions. SE in brackets account for plot differences.

Life Form		3 months		6 months	
		No Seedlings	% of Total	No Seedlings	% of Total
Native	Grass	105 (± 2.0)	12 %	150 (± 5.5)	12 %
	Sedge	16 (± 1.6)	2 %	29 (± 2.7)	2 %
	Herb	640 (± 46.4)	74 %	980 (± 62)	77 %
	Woody Perennial	15 (± 1.8)	2 %	18 (± 2.0)	1 %
Non-native		92 (± 4.5)	10 %	104 (± 5.0)	8 %
	Total	868 (± 57.3)		1281 (± 62.2)	

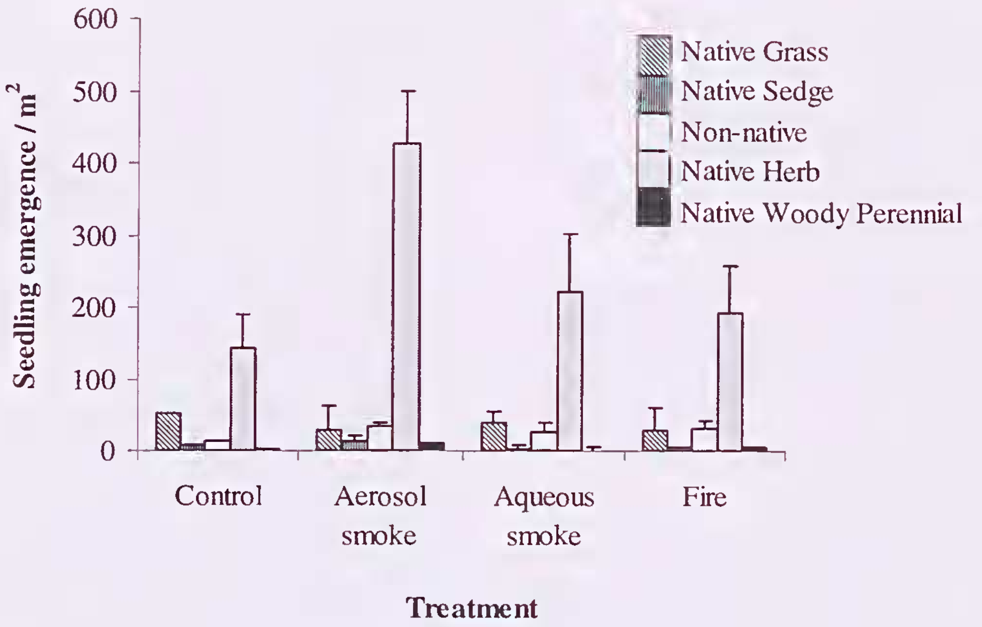

Figure 2. Seedling emergence by treatment for each life form 6 months after sowing.

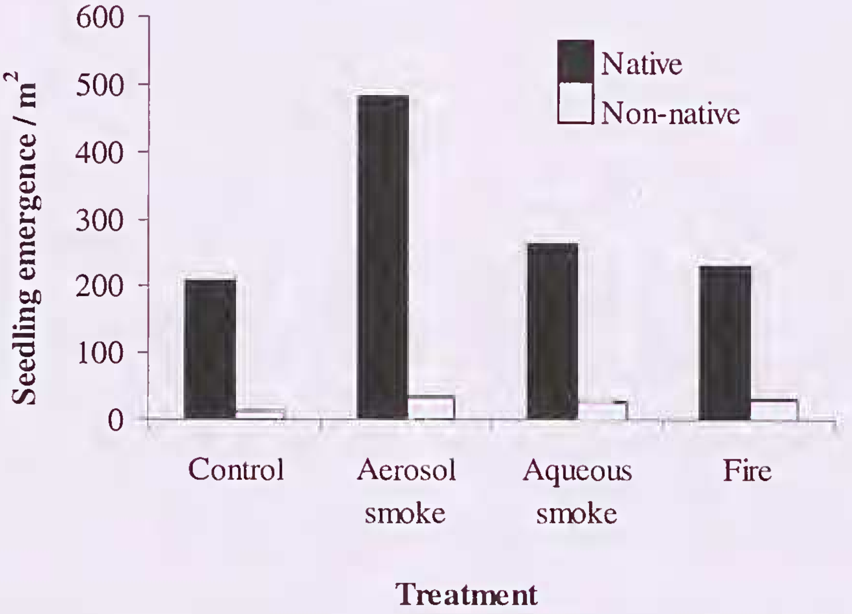

Figure 3. Relationship between native and non-native seedlings emerging by treatment after six months under shade house conditions.

Discussion

The level of seedling emergence in this study (256 seedlings / m^2) was similar to that reported by Enright *et al.* (2004) for the crest and swale shrublands near Eneabba, south western Australian, low compared to the 767 seedlings / m^2 that Vlahos & Bell (1986) reported from the northern jarrah forest of Western Australia, but higher than the germinable soil seed store reported by Bellairs & Bell (1993) (140–174 seedlings / m^2) from the kwongan vegetation north of Perth. The differences in seedling emergence between these investigations may be reflecting site and species differences, the longevity of the soil seed bank at each site and its post-fire age. Seed of many species may be found in the soil for only a relatively short period of time following shedding, showing seasonal peaks of abundance (Thompson 1992) and seasonal heterogeneity in soil seed bank samples was reported by Koch *et al.* (1987) from a Western Australian jarrah (*Eucalyptus marginata*) forest. Conducting investigations into the time of sampling the soil seed bank may be important for those species that seed intermittently, and sampling throughout a season should take into account natural seed mortality in the soil. The post-fire age of a site can also influence the composition of the seed bank, with early successional plants tending to produce dormant seeds (e.g., acacias) and late successional species tending to lose viability quickly (Bazzaz 1979).

Vegetation communities dominated by perennial species with canopy-borne seed (*e.g., Banksia, Dryandra, Allocasuarina, Hakea*), or species with low seed production and /or capable of resprouting (*e.g., Xanthorrhoea*), may not show a good relationship between above- and below-ground species composition. Many Australian species store their seed in the canopy (Bell *et al.* 1993) and only release seed when fruits are opened by fire (Lamont *et al.* 1991). Once released, these canopy-borne seed generally have no dormancy mechanisms and germinate readily (Bell *et al.* 1993). The seed of these species will be largely absent in the soil seed reserve, as demonstrated in this study. Thompson (1979), Vlahos & Bell (1986) and Wills & Read (2002) also reported the low incidence of seed of major woody tree and shrub species emerging from the soil seed bank. At the study site, soil contained seeds of many herbaceous species absent from the vegetation, providing a poor correlation between the standing vegetation and the species present in the soil seed bank at the time of sampling. Particular disturbance treatments provided to seeds in this study appear to have stimulated germination in a range of otherwise dormant herbaceous species. For instance, the seed of post-fire colonizers may be represented in the soil seed reserve, but likely to be missing from the standing vegetation in long unburnt vegetation communities. This hidden component of the vegetation community is often made up of ephemerals. The emergence of the post-fire native herb *Trachymene*, absent from the extant vegetation at East Yornaning, was evidence of this.

Our results showed that specific treatments enhanced germination of different life forms, increased the species richness of different components of the seed bank, and had different effects on the rate of emergence. More than one third of all seedlings that emerged within six months from the soil seed bank appeared after aerosol smoke application, whereas less than one quarter of soil-stored seeds were stimulated to germinate after aqueous smoke treatment. Aerosol smoke application has been shown to significantly enhance germination of many Western Australian plant species (Dixon *et al.* 1995; Lloyd *et al.* 2000) with some evidence that aerosol smoke application is superior to aqueous smoke for stimulation of germination of a wide range of seed types (Roche *et al.* 1997). Grasses were the only life form not stimulated by aerosol smoke application in our study, contrasting with results reported by Read & Bellairs (1999) and Read *et al.* (2000), who noted a significant stimulatory effect of smoke on the germination of native grasses on the east coast of Australia. Unfortunately, aerosol smoke application also favourably affected germination of non-native species at the study site, a response previously noted by de Lange & Boucher (1990) and Jager *et al.* (1996) in South Africa and Adkins *et al.* (2000) in Australia. Read *et al.* (2000) reported that non-native species contributed to some 20% of the seed bank in untreated topsoil from a dry sclerophyll forest in the Hunter Valley, New South Wales and cautioned the possible negative effect of smoke in creating a future weed problem in native ecosystems.

Fire plays an important role in influencing seed germination in plant communities (Bell 1999). Fire can influence the composition and density of species and provides a thermal shock, which acts to break dormancy in many species with physical dormancy (Bell 1999). The fire treatment in this study was designed to provide heat-requiring seed with sufficient heat to break dormancy, but the low fuel levels in the fire treatment may have contributed to low emergence levels of hard seeded perennial species. Unfortunately, fire can also assist the incursion of non-native plant species into natural areas (Abensperg-Traun *et al.* 1998, Drake 1998). Non-native species were found to be more abundant in the soil seed bank than in the standing vegetation at the study site. The presence of non-native species in the soil-stored seed reserve in a relatively weed-free environment can indicate either a historical presence of these species or long distance seed dispersion taking place. *Hypochaeris, Sonchus, Arctotheca* and *Ursinia* are non-competitive non-native plants that are wind dispersed and their presence does not indicate ecosystem degradation *per se*. Some of these non-native species may have extremely long lived soil-stored seed that accumulates in the soil over many years, suggesting an increased frequency of non-native plant species in the standing vegetation in the past. Enright *et al.* (1997) also reported a seed reserve of non-native species despite absence of these species in the standing vegetation in a large National Park in south eastern Australia.

The success of native regeneration post-disturbance is largely dependent on the post-disturbance environment. Competition from non-native plants may have an effect on the composition of regenerating vegetation. For example, Perez-Fernandez *et al.* (2000) reported the superior germination ability of non-native species over native species in southwest Australia attributing this response to the formers' ability to utilize moisture more rapidly. Distance from a reserve edge may affect the density of non-native species, with fewer non-natives identified at increasing distances from a reserve boundary (Hester & Hobbs 1992). On the other hand, weed seed presence in the soil is strongly influenced by the availability of seed dispersal vectors. Long distance dispersal, by wind or through animal dispersion, may explain the presence of non-native plant seed within a reserve (Hussey *et al.* 1997). At East Yornaning, the seeds of most of the non-native species that emerged from the soil-stored seed bank (e.g., members of the Asteraceae family) have mechanisms for wind dispersal.

At present there is little known about the response of Western Australia's southern wheatbelt woodland vegetation to disturbance. This investigation suggests that smoke treatment provides a more effective stimulant for the soil-stored seed bank than the heat and smoke generated by fire for a range of life forms. Despite little evidence of invasive weeds in the standing vegetation, the incidence of seeds of non-native herbs and grasses emerging from the seed bank may be of concern to reserve management in the event of wildfire. The conditions favourable for establishment would be dependant on type of disturbance and subsequent rainfall events. These weed species have not been yet identified as potential problems, although their incursion into remnant reserves in agricultural landscapes should be closely monitored. Although not a concern for reserve management at the present time, it is of interest to note that non-native plant species may be capable of invading remnant vegetation like East Yornaning Nature Reserve in the future.

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